In vivo X-ray elemental imaging of single cell model organisms manipulated by laser-based optical tweezers

E. Vergucht[1], T. Brans[2,3], F. Beunis[2,3], J. Garrevoet[1], M. De Rijcke[4], S. Bauters[1,5], D. Deruytter[4], M. Vandegehuchte[4], I. Van Nieuwenhove[6], C. Janssen[4], M. Burghammer[5,1], L. Vincze[1]

[1]X-ray Microspectroscopy and Imaging Group, Ghent University, Ghent, Belgium (**eva.vergucht@ugent.be**)

[2]Department of Electronics and Information Systems, Ghent University, Ghent, Belgium

[3]Center for Nano and Biophotonics, Ghent University, Ghent, Belgium

[4]Laboratory of Environmental Toxicology and Aquatic Ecology, Ghent University, Ghent, Belgium

[5]European Synchrotron Radiation Facility, Grenoble, France

[6]Polymer Chemistry and Biomaterials Group, Ghent University, Ghent, Belgium

We report on a radically new elemental imaging approach for the analysis of biological model organisms and single cells in their natural, *in vivo* state. The methodology combines optical tweezers (OT) technology for non-contact, laser-based sample manipulation with synchrotron radiation confocal X-ray fluorescence (XRF) microimaging *for the first time*. The main objective of this work is to establish a new method for *in situ* elemental imaging of free-standing living biological microorganisms or single cells in their aqueous environment. Using the model organism *Scrippsiella trochoidea*, a first proof of principle experiment at ESRF Microfocus beamline ID13 demonstrated the feasibility of the OT XRF methodology. In a second experiment, the OT XRF methodology was successfully combined with complementary integrated SAXS imaging to visualize the sample outline. Moreover, we studied the mixture toxicity of Cu-Ni and Cu-Zn as a result of elevated exposure and semi-quantitatively observed rather large differences in the *in vivo* areal concentrations of accumulated metals in single cells.

We expect that the OT XRF methodology will significantly contribute to the new trend of investigating microorganisms at the cellular level with added *in vivo* capability [1,2].

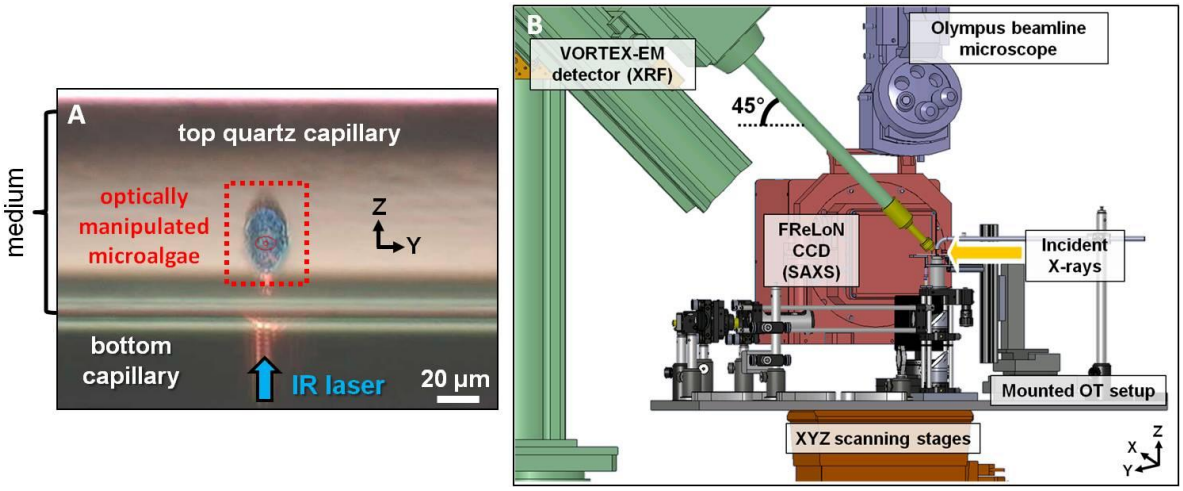

Figure 1: (a) Optically manipulated *S. trochoidea* microalgae in aqueous medium. (b) OT XRF methodology integrated at ESRF-ID13, OT setup mounted on scanning stages, XRF detector positioned under 45° and equipped with a confocal optic, FReLoN CCD camera positioned downstream for integrated SAXS imaging.

References

[1]-S.C. Santucci, D. Cojoc, H. Amenitsch, B. Marmiroli, B. Sartori, M. Burghammer, S. Schoeder, E. DiCola, M. Reynolds, C. Riekel, Analytical Chemistry **83**, 4863–4870 (2011).

[2]-E. Vergucht, T. Brans, F. Beunis, J. Garrevoet, M. De Rijcke, S. Bauters, D. Deruytter, M. Vandegehuchte, I. Van Nieuwenhove, C. Janssen, M. Burghammer, L. Vincze, Submitted to Scientific Reports (2014).